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## Determination of Yielding Ability, Free Radical Scavenging Activity, Protein and Carotenoid Contents of Selected Genotypes of *Amaranthus tricolor*

P. Malathy<sup>a,\*</sup>, D.D Suraweera<sup>b</sup>, W. A. M. Daundasekara<sup>b</sup>, W.D.G.P. Nilanthi<sup>a</sup>,  
K.B.Wahundeniya<sup>a</sup>

<sup>a</sup>Horticultural Crops Research and Development Institute, Department of Agriculture, Gannoruwa, Peradeniya, Sri Lanka, 20400<sup>b</sup>Department of Botany, University of Peradeniya, Peradeniya, Kandy, 20400

### Abstract

Identification of new leafy vegetable varieties with high yield potential and nutritional components is one of the priority areas of the Department of Agriculture, Sri Lanka. As a part of this activity, yield potential, nutritional values and antioxidant properties of promising accessions of *Amaranthus tricolor* were studied with the aim of selecting better varieties. A series of experiments were carried out using three new genotypes namely *DOA red*, *Pure green* and *Diyapalagoda* along with the recommended variety, *DOA green*. Yield potential was determined by evaluating growth and yield parameters. Comparison of protein levels of harvested leaves was done by Kjeldahl method. Carotenoid, anthocyanin, chlorophyll levels and the antioxidant activity of the leaves of four genotypes at harvest was determined based on UV-spectrophotometric method. Genotype *Pure green* exhibited good growth and yield performance, wide adaptability and higher amount of carotenoids. Hence it can be recommended for commercial cultivation. The faster growth of genotype *Diyapalagoda* indicated its suitability for home garden cultivation. *DOA red* exhibited higher amount of chlorophylls, and anthocyanin and the greatest antioxidant activity. *DOA green* contains the highest percentage of crude protein.

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**Keywords:** Amaranthus, antioxidant, leafy vegetables

\* Corresponding author. Tel.: 0094812388011-2; fax: 00948123882334.  
E-mail address: [mpmalathy@yahoo.com](mailto:mpmalathy@yahoo.com).

## 1. Introduction

Daily dietary intake of leafy vegetables would help to reduce the malnutrition problem since they provide essential minerals, vitamins and amino acids that are absent in the rice based diet. They are less expensive and easily found in any part of this country compared to other vegetables. *Amaranthus tricolor* (Amaranth), is one of the popular leafy vegetables in Sri Lanka due to its easiness in culture, fast growth rate, adaptability to varying agro climates and high yield potential. *A. tricolor* leaves are reported to contain high amount of crude protein and nutritionally critical amino acids viz. lysine and methionine (Oliveira and de Carvalho, 1975) in addition to dietary fiber, vitamins and dietary minerals (Oke, 1983). It is reported to contain high amount of antioxidant compounds including vitamin C, carotenoids and phenolic compounds such as flavonoids (Romani *et al.*, 2002).

In Sri Lanka, different selections of Amaranth are being cultivated by farmers, but very little information is available on their growth and yield performance, nutritional value and quality characters. This may lead to cultivation of poor quality selections with low yield (Wahundeniya, 2009). Hence, several accessions of Amaranth have been collected from farmer fields, purified and subjected to yield and quality evaluation at Horticultural Crops Research and Development Institute (HORDI). To identify varieties with best adaptation to any particular location, it is important to compare the yield potential during different growing seasons. For the selection of superior variety, evaluation of antioxidant activity can be used as a relatively novel index together with morphological characterization, and yield evaluation (Ogunlesi *et al.*, 2003). Therefore, yield potential, some nutritional aspects and anti-oxidant properties of promising accessions of *A. tricolor* were studied with the aim of selecting better varieties for commercial cultivation.

## 2. Materials and Methods

Experiments were conducted at HORDI during two consecutive years using four selected genotypes, *DOA red*, *Pure green*, *Diyapalagoda* and *DOA green* (control variety) of *A. tricolor* to study their growth and yield performance, and nutritional values with special reference to protein level, plant pigment content and antioxidant activity in leaves. These experiments were laid out in a Randomized Complete Block Design with four replicates. Agronomic practices were adopted according to DOA recommendation. Growth parameters viz. plant height (cm), number of leaves, stem circumference (cm), length of 3<sup>rd</sup> internode (cm), width (cm) and length of 6<sup>th</sup> leaf (cm) were measured in addition to fresh weight per plant (g) and leaf, stem and root weight in wet and dry basis and total yield (mt/ha).

### 2.1. Determination of the amount of crude protein in *A. tricolor* leaves

The crude protein content in leaves was determined using Kjeldahl method (Cunniff, 1998).

### 2.2. Determination of carotenoid, chlorophyll and anthocyanin levels of *A. tricolor* leaves

Absorbance of leaf extract at 470nm, 537nm, 647nm and 663nm was measured using spectrophotometer (UV 1800) (Lichtenthaler *et al.*, 1983). Concentration of carotenoids, chlorophylls and anthocyanin were calculated according to Daniel *et al.*, (2002).

### 2.3. Determination of the antioxidant activity (free radical scavenging activity) of *A. tricolor* leaves

This was analyzed with slight modifications to the method reported by Patel *et al.*, (2010).

Data were subjected to statistical analysis by ANOVA using SAS software package (1998). Mean separation was done using LSD at  $p < 0.05$  level.

### 3. Results

#### 3.1. Comparison of the growth parameters of four genotypes of *A. tricolor*

Among the growth parameters tested, plant height, number of leaves, weight of fresh leaf and plant, fresh and dry weight of stem and roots and total yield differed significantly (Table 1). The genotype, *Pure green* showed significantly high yield throughout the tested period. There was no genotype in to environment interaction observed in *Pure green* (Table 2).

#### 3.2. Crude protein, Carotenoid, chlorophylls and anthocyanin levels of *A. tricolor* leaves

The genotype *DOA green* leaves had the highest amount of crude protein and the genotype *Pure green* contained the lowest amount of crude protein (Table 3). There was a significant variation observed in pigment contents, among the genotypes (Table 3). The genotype *DOA red* exhibited higher amount of chlorophylls and anthocyanin while higher amount of carotenoids was recorded in *Pure green*.

#### 3.3. Antioxidant activity of leaves in four genotypes of *A. tricolor*

Among the four *A. tricolor* genotypes studied, *DOA red* exhibited the greatest DPPH free radical scavenging activity as indicated by the lowest  $IC_{50}$  value whereas *DOA green* showed the lowest (Table 4).

Table 1. Growth performance of four genotypes of *A. tricolor*

Parameter	Genotype			
	<i>DOA red</i>	<i>DOA green</i>	<i>Pure green</i>	<i>Diyapalagoda</i>
Plant height (cm)	65.90 <sup>b</sup>	65.44 <sup>b</sup>	70.39 <sup>b</sup>	77.68 <sup>a</sup>
Stem circumference (cm)	3.79	3.78	3.80	23.47
Number of leaves	38.90 <sup>c</sup>	43.85 <sup>c</sup>	85.58 <sup>a</sup>	53.05 <sup>b</sup>
Length of 6th leaf (cm)	20.18	19.39	19.61	19.80
Width of 6th leaf (cm)	13.86	13.14	13.07	15.78
3rd intermodal length (cm)	2.99	3.31	3.71	3.30
Fresh weight per plant (g)	174.77 <sup>b</sup>	188.06 <sup>b</sup>	281.25 <sup>a</sup>	278.00 <sup>a</sup>
Fresh leaf weight (g)	62.89 <sup>b</sup>	63.68 <sup>b</sup>	96.36 <sup>a</sup>	84.29 <sup>a</sup>
Dry leaf weight (g)	6.38	6.74	8.07	7.53
Fresh stem weight (g)	71.18 <sup>b</sup>	77.47 <sup>b</sup>	120.15 <sup>a</sup>	126.09 <sup>a</sup>
Dry stem weight (g)	3.70 <sup>b</sup>	4.02 <sup>b</sup>	6.26 <sup>a</sup>	6.60 <sup>a</sup>
Fresh root weight (g)	16.47 <sup>b</sup>	13.66 <sup>b</sup>	33.81 <sup>a</sup>	35.76 <sup>a</sup>
Dry root weight (g)	1.57 <sup>cb</sup>	1.37 <sup>c</sup>	2.37 <sup>a</sup>	2.89 <sup>a</sup>

Mean values followed by different letters in the column are significantly different at  $P \geq 0.05$  level

Table 2. Yielding ability of four genotypes of *A. tricolor* in four different seasons

Variety	Yield (mt/ ha)			
	Season 1	Season 2	Season 3	Season 4
<i>DOA red</i>	17.64 <sup>c2</sup>	21.44 <sup>c1</sup>	19.13 <sup>b2</sup>	16.42 <sup>c2</sup>
<i>DOA green</i>	22.48 <sup>b1</sup>	24.76 <sup>c1</sup>	11.89 <sup>b3</sup>	21.93 <sup>b1</sup>
<i>Pure green</i>	32.24 <sup>a1</sup>	36.72 <sup>a1</sup>	27.93 <sup>a1</sup>	34.33 <sup>a1</sup>
<i>Diyapalagoda</i>	22.09 <sup>b2</sup>	31.01 <sup>ab1</sup>	22.30 <sup>ab2</sup>	24.46 <sup>b2</sup>

Mean values followed by different letters in the rows and numbers in the columns are significantly different at  $P \geq 0.05$  level

Table 3: Amount of crude protein levels, carotenoids, chlorophylls and anthocyanin levels in the four selected genotypes of *A. tricolor*

Genotype	% crude protein	Anthocyanin	Chlorophyll a	Chlorophyll b	Carotenoids
		$\mu\text{g}/\text{cm}^2$	$\mu\text{g}/\text{cm}^2$	$\mu\text{g}/\text{cm}^2$	$\mu\text{g}/\text{cm}^2$
<i>DOA red</i>	23.63 <sup>ab</sup>	1.738 <sup>a</sup>	9.796 <sup>e</sup>	3.075 <sup>i</sup>	3.846 <sup>xy</sup>
<i>DOA green</i>	24.8 <sup>a</sup>	0.723 <sup>a</sup>	2.223 <sup>f</sup>	2.816 <sup>ij</sup>	4.815 <sup>x</sup>
<i>Pure green</i>	22.8 <sup>b</sup>	1.224 <sup>a</sup>	2.376 <sup>f</sup>	2.983 <sup>ij</sup>	5.155 <sup>x</sup>
<i>Diyapalagoda</i>	23.43b	1.375 <sup>a</sup>	8.896 <sup>ef</sup>	2.461 <sup>j</sup>	3.628 <sup>y</sup>

Mean values followed by different letters in each row are significantly different at  $P \geq 0.05$  level

Table 4. Comparison of the DPPH radical scavenging activity of four genotypes of *A. tricolor* ethanolic extracts and standard ascorbic acid

Sample	Equation	R <sup>2</sup>	IC <sub>50</sub> values ( $\mu\text{g}/\text{ml}$ )
Standard Ascorbic acid	$y = 5.437x + 31.01$	0.996	34.92
<i>DOA red</i>	$y = 0.026x + 37.18$	0.997	493.07
<i>DOA green</i>	$y = 0.021x + 25.60$	0.994	1161.91
<i>Pure green</i>	$y = 0.014x + 35.72$	0.992	1020.00
<i>Diyapalagoda</i>	$y = 0.020x + 35.57$	0.991	721.50

IC<sub>50</sub> value denotes the 50% inhibition concentration

#### 4. Discussion

Yield potential is determined by genetic makeup of the plant and environmental factors (Malik *et al.*, 2009). Among the tested genotypes *Pure green* exhibited best growth performance and yielding ability. The highest leaf to stem ratio was also recorded for the same genotype (data not shown), which enhances the marketable value and consumer preference. Furthermore, absence of genotype X environment interactions over four consecutive seasons evidenced the wider adaptability of *Pure green*. The genotype *Diyapalagoda* exhibited highest plant height and root weight. Higher growth rate favors several harvests in a season and well developed root system provides the ability to grow under wide range of climatic conditions. The growth performance observed in genotype *Diyapalagoda* indicated its suitability for home garden cultivation.

Higher protein varieties tend to have lower yields and vice versa (McKenzie *et al.*, 2008). Carotenoids,

ascorbic acid, flavonoids and phenolic compounds are some of the components which contribute to high antioxidant activity. Among the genotypes tested, *DOA red* exhibited the highest antioxidant activity and this might be due to its significantly higher total pigment content compared to that in other genotypes. Anthocyanins in red colour of vegetables are reported to contribute significantly to antioxidant activity (Velioglu *et al.*, 1998). Carotenoids also contribute significantly to total antioxidant capacity of leafy vegetables through their ability to neutralize free radicals by donor electron, without becoming active free radicals (Mortensen *et al.*, 1997). Chlorophyll and its derivatives are also known to have antioxidant activity. The results of the present study agree with the fact that the total contents of above pigments have a direct relationship with the antioxidant activity.

## 5. Conclusions

Genotype *Pure green* exhibited good growth and yield performance, wide adaptability and higher amount of carotenoids. Hence it can be recommended for commercial cultivation. The faster growth of genotype *Diyapalagoda* indicated its suitability for home garden cultivation. *DOA red* exhibited the greatest antioxidant activity, higher amount of chlorophylls, and anthocyanin. *DOA green* contains the highest percentage of crude protein.

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